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# Screening of the Chickpea germplasm for resistance to *Sclerotium rolfsii* Sacc. Incitant of collar rot disease

**Jasmin Thomas, R Sarada Jayalakshmi Devi, S Khayum Ahammed, V Jayalakshmi and VLN Reddy**

**Abstract**
Chickpea collar rot caused by *Sclerotium rolfsii* is considered economically important soil borne disease. A total of 20 Chickpea advanced breeding lines supplied by Chickpea Breeder, RARS, Nandyal were screened under field conditions (Screening blocks) against collar rot. L-550 was used as a susceptible check. The observations *viz.*, total number of plants and total number of infected plants were recorded up to up to 30 days after sowing and Percent disease incidence of collar rot was calculated. Among 20 advanced breeding lines, 6 lines (NBeG 1267, NBeG 440, NBeG 690, NBeG 779, NBeG 699 and NBeG 776) were showed resistant reactions with 0-10 per cent disease incidence, 10 lines were found to be moderately resistant reactions, 2 lines were reacted as moderately susceptible reaction, 2 lines were showed susceptible reaction and L 550 showed highly susceptible reactions.

**Keywords:** Chickpea, genotypes, screening, resistance

## Introduction
Chickpea (*Cicer arietinum* L.) is one of the most cultivated legume crops and a rich source of protein in many countries. It is rich in energy, vitamins, protein, minerals, fiber, and beneficial phytochemicals for health (Wood and Grusak, 2007; Jukanti *et al.*, 2012) [18, 6]. Diseases of chickpea considered as the major constraint for improvement of the crop yield. Three soil borne diseases such as wilt, collar rot and dry root rot are considered economically important. Among these, collar rot caused by *Sclerotium rolfsii* has an important role which about 10–30% yield loss has been observed annually (Maurya, 2008) [10]. Control of *S. rolfsii* has been extremely difficult due to the pathogen's prolific growth, wide host range, and ability to produce a large number of sclerotia that can persist in the soil for several years (Sennoi *et al*., 2013) [16]. It has wide host range and attack over 200 different species causing diseases on a wide range of agricultural and horticultural crops (Parvin *et al*. 2016; Billah, 2017) [12, 2]. The disease symptoms mainly occur in wet soil conditions, which appear within two weeks of sowing and the foliage turns yellow before drying and the death of the plant. The collar region shows the rotting symptom and the rotted area covers with white mycelial strands of *S. rolfsii* with mustard like sclerotia around the infected portion of root (Lahre, 2008; Khan *et al.*, 2020) [8, 7]. The development of disease resistant cultivars is the most practicable and cost-effective control strategy for such a devastating soil-borne pathogen (Akram *et al.*, 2008) [1]. The accurate simulation of natural environmental conditions where plants are exposed to the inoculum is required for effective disease resistance screening (Choudhary *et al.*, 2013) [3]. The present investigation was conducted to screen the chickpea advanced breeding lines against *S. rolfsii* for the identification of resistant sources in available genotypes.

## Materials and Methods
### Isolation of the pathogen from infected plant samples
Tissue segment method (Rangaswami and Mahadevan, 1999) [14] was followed for isolation of pathogen. Infected collar portion along with some healthy portions made cut with the help of razor blade under aseptic conditions. Surface sterilization with 1.0 percent sodium hypochlorite solution for one minute was followed by three washes with sterile distilled water to remove any traces of sodium hypochlorite. These bits were aseptically placed to petriplates containing sterilized PDA media after being transferred to sterile blotting paper to remove water adhered to the sample. The plates were incubated in incubator at 27±2 °C and observed periodically for growth of the fungus. After attaining fungal growth, small disc measuring 5 mm was cut and transferred aseptically to the PDA slants to obtain the pure culture of the fungus.

**Mass multiplication of pathogen**

Sorghum grains were selected for mass multiplication of pathogen. Sorghum grains were soaked in 2% sucrose solution overnight, next day boiled in fresh water for 30 minutes and drained to remove excess water. They were transferred to 250 ml conical flasks @ 100g and autoclaved for 15 p.s.i at 121.6$^0$C for 15 minutes. The flasks were cooled at room temperature and then inoculate 5 mm discs of 3 to 4 day old culture of *S. rolfsii* grown on PDA to conical flasks under aseptic conditions and kept for incubation at 27 ± 2$^0$C for two weeks. The flasks were agitated regularly to obtain a uniform growth all over the flasks. After 2 weeks the flasks were used for inoculating soil in sick plot.

**Screening of chickpea entries**

A total of 20 Chickpea advanced breeding lines supplied by Chickpea Breeder, RARS, Nandyal were screened under field conditions (Screening blocks) against collar rot during *Rabi* 2021. *S. rolfsii* multiplied on sorghum grains were added to soil and mixed thoroughly. The field experiments were laid out in a randomized block design with two replications. Each line was sown in 3 m row length with 30 x10 cm$^2$ spacing. After every five test genotypes one line of L-550 susceptible check was sown. The observations *viz.*, total number of plants and total number of infected plants were recorded up to up to 30 days after sowing and Percent disease incidence of collar rot was calculated. PDI was calculated by using the following formula.

$$\text{Per cent disease incidence} = \frac{\text{Number of plants infected}}{\text{Total number of plants}} \times 100$$

Based on disease incidence the genotypes were categorized into different groups as given in Table 1.

**Table 1:** Table showing disease incidence scale

| Reaction | Collar rot incidence (%) |
|---|---|
| Resistant | 0 – 10 |
| Moderately resistant | 11 – 20 |
| Moderately susceptible | 21 – 30 |
| Susceptible | 31-50 |
| Highly susceptible | >50 |

***Source*:** AICRP chickpea 2018 proceedings

**Results and Discussion**

Among 20 advanced breeding lines 6 lines (NBeG 1267, NBeG 440, NBeG 690, NBeG 779, NBeG 699 and NBeG 776) were showed resistant reactions with 0-10 per cent disease incidence, 10 lines (NBeG 452, NBeG 844, NBeG 789, KAK 2, NBeG 798, NBeG 47, NBeG 924, NBeG 1137, NBeG 810 and NBeG 934) were found to be moderately resistant reactions with 11-20 per cent disease incidence, 2 lines (NBeG 1146 and NBeG 506) were reacted as moderately susceptible reaction with 21-30 per cent disease incidence, 2 lines (NBeG 857 and NBeG 833) were showed susceptible reaction with 30-50 per cent disease and L 550 showed highly susceptible reactions (Table 2, Table 3, Fig 1).

**Table 2:** Screening of chickpea advanced breeding lines against collar rot

| Advanced breeding line | Type | Germination (%) | Per cent disease incidence | Reaction |
|---|---|---|---|---|
| NBeG 1267 | Desi | 100 | 3.33 | Resistant |
| NBeG 440 | Kabuli | 95 | 3.50 | Resistant |
| NBeG 452 | Desi | 90 | 18.51 | Moderately resistant |
| NBeG 506 | Desi | 83.3 | 24.00 | Moderately susceptible |
| NBeG 844 | Kabuli | 96.6 | 13.79 | Moderately resistant |
| NBeG 789 | Kabuli | 86.6 | 11.58 | Moderately resistant |
| NBeG 857 | Desi | 75 | 31.11 | Susceptible |
| KAK 2 | Kabuli | 93.3 | 17.85 | Moderately resistant |
| NBeG 690 | Desi | 96.6 | 3.44 | Resistant |
| NBeG 798 | Desi | 100 | 16.66 | Moderately resistant |
| NBeG 779 | Desi | 95 | 7.01 | Resistant |
| NBeG 699 | Desi | 100 | 6.66 | Resistant |
| NBeG 924 | Desi | 78.3 | 12.76 | Moderately resistant |
| NBeG 47 | Desi | 80 | 12.50 | Moderately resistant |
| NBeG 1137 | Desi | 85 | 11.76 | Moderately resistant |
| NBeG 776 | Desi | 100 | 6.66 | Resistant |
| NBeG 833 | Kabuli | 90 | 30.03 | susceptible |
| NBeG 810 | Kabuli | 85 | 11.76 | Moderately resistant |
| NBeG 934 | Kabuli | 93.3 | 14.28 | Moderately resistant |
| NBeG 1146 | Desi | 86.6 | 21.15 | Moderately susceptible |
| L-550 | Desi | 90 | 51.85 | Highly susceptible |
| C.D. | | 10.61 | 6.80 | |
| SE(m) | | 3.57 | 2.29 | |
| SE(d) | | 5.05 | 3.24 | |
| C.V. | | 5.56 | 24.80 | |

**Table 3:** Reaction of chickpea entries against collar rot

| S. No. | Disease Reaction | Advanced breeding lines | No. of AB lines |
|---|---|---|---|
| 1 | Resistant (0-10% disease incidence) | NBeG 1267, NBeG 440, NBeG 690, NBeG 779, NBeG 699, NBeG 776 | 6 |
| 2 | Moderately resistant (11-20% disease incidence) | NBeG 452, NBeG 844, NBeG 789, KAK 2, NBeG 798, NBeG 47, NBeG 924, NBeG 1137, NBeG 934, NBeG 810 | 10 |
| 3 | Moderately susceptible (21-30% disease incidence) | NBeG 1146, NBeG 506 | 2 |
| 4 | Susceptible (31-50% disease incidence) | NBeG 857, NBeG 833 | 2 |
| 5 | Highly susceptible (>50% disease incidence) | L 550 | 1 |

Six genotypes were categorised as resistant as they showed a disease incidence less than 10%. These genotypes can be exploited as resistant sources in breeding programmes. The advanced breeding lines that showed resistance reaction against collar rot in the present study were also identified as resistant against Fusarium wilt (Manasa *et al*., 2020) [9]. Similarly, Noor (2019) [11] screened 16 advanced breeding lines in screening block against collar rot and found 2 resistant advanced breeding lines namely, NBeG 699 and NBeG 810 with PDI 10% and 0.00% respectively. In contrast, in the present study, NBeG 810 was found be moderately resistant. Due to the breakdown of resistance, it is necessary to continuously screen the germplasm of chickpea for resistance against soil-borne diseases.

The present results are in agreement with Gupta and Mishra (2009) [4] who screened 120 lines of chickpea in disease sick fields for 3 consecutive years and 32 entries performed consistent resistant reaction to collar rot. Twelve accessions were found free from collar rot during the testing years under high disease pressure. Hassan *et al.* (2012) [5] screened 116 chickpea lines under field conditions against collar rot disease. There were 33 genotypes with a resistant reaction, 33 with a moderately resistant reaction, 38 with a moderately susceptible reaction, and 12 with a susceptible reaction.

Ramesh *et al.* (2014) [13] screened 88 desi and 11 kabuli chickpea genotypes in pot house against *S. rolfsii*. Among desi genotypes GNG 1958 was found to be resistant to disease whereas, 13 entries were moderately resistant. Among kabuli types, 2 entries i.e. GNG 1969, BG 2086 were resistant and 9 as moderately resistant. Shirsole *et al.* (2018) [17] screened 185 chickpea entries under field condition against *S. rolfsii*. Among them 5 entries exhibited moderate resistance while, the remaining were susceptible to highly susceptible for collar rot of chickpea.

Cultivation of resistant varieties is an important cost effective strategy for the management of soil-borne diseases in chickpea (Sarwar *et al.*, 2012) [15]. An understanding of the genetic diversity of the pathogen and its environment is an important prerequisite in developing and deploying varieties.

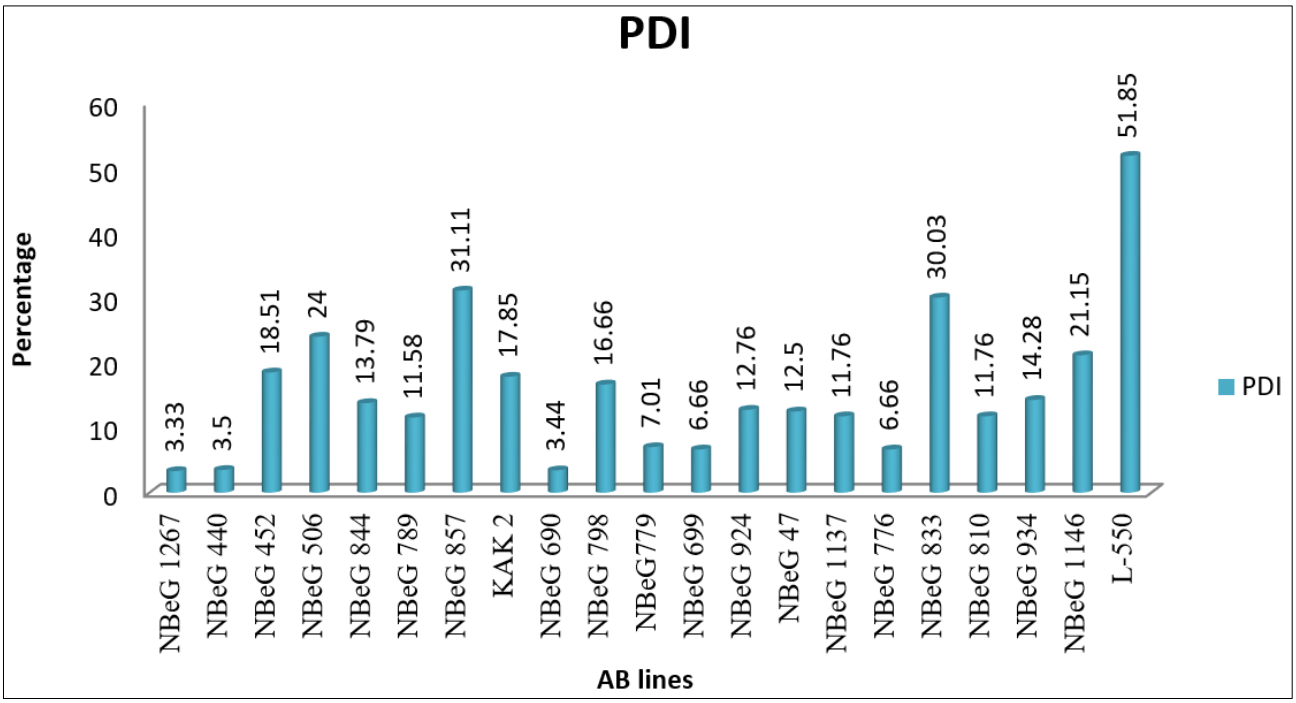


**Fig 1:** Screening of chickpea advanced breeding lines against collar rot.

## Conclusion

Out of 20 advanced breeding lines screened in the screening block, 6 lines showed resistant reactions, 10 lines showed moderately resistant reactions, 2 lines were found to be moderately susceptible, and 2 lines were recorded as susceptible in comparison with the highly susceptible check. The utilization of resistant varieties is an economical approach disease management practice for such a devastating soil-borne pathogen.

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